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HEMOLYTIC STREPTOCOCCI IN THE THROAT OF THE DOG

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In view of the interest in hemolytic streptococci it seemed desirable to study the prevalence and characteristics of such streptococci in the throat of the dog.

Cultures were made by swabbing the throats of live dogs, endeavoring particularly to reach the tonsils and nasopharynx, and inoculating aerobic 10% goat blood-agar plates. Hemolytic colonies were transferred to plain broth, from the broth cultures blood agar plates made, and the single hemolytic colonies studied further. The hemolytic colonies on the plates were all of the beta type of hemolysis; that is, the hemolytic zone surrounding the colony was from 2-4 mm. in diameter and the margin was sharp and distinct. Only those cocci were chosen for further study that grew into long chains in broth. A total of 21 strains was thus secured. Stock cultures were made on plain blood-agar slants. Typical hemolytic streptococci were isolated from the throat in 39% of 32 dogs examined.

The chains in broth cultures varied from 15-100 cocci. The individual cocci were spherical or slightly flattened at the adjoining poles. In most of the smears, some chains contained large, irregular, swollen cocci. No capsules were observed.

Sixteen of the 21 strains were pathogenic for mice when 1 c c of an 18-hour broth culture was inoculated intraperitoneally; the mice dying within 24 hours.

All the 21 strains brought about complete laking when 0.5 c c of broth culture was transferred to 0.5 c c of a 5% suspension of washed rabbit erythrocytes and the mixtures incubated for 2 hours at 37 C. in a water-bath.

For bile solubility, 0.2 c c of ox bile were added to 1 c c of broth culture and observed after one hour incubation at 37 C. None of the 21 strains were soluble in bile by macroscopic and microscopic observation.

The growth of the streptococci was scanty in plain broth; there was a thin, granular, somewhat adherent sediment that in the majority

of cases extended up one side of the tube. The supernatant fluid was clear.

The fermentative reactions were determined in Hiss sheep serum medium, containing, respectively, 1% of dextrose, inulin, lactose, maltose, mannite, raffinose, saccharose, salicin and starch; also the plain medium without added carbohydrate. The results in the sugar mediums are given in table 1. There were no changes evident in the inulin and plain mediums. There was no gas produced in any sugar.

TABLE 1
FERMENTATIVES REACTIONS IN SUGAR MEDIUMS

No. of Strain	Litmus Milk	Carbohydrates							
		Dex-trose	Lac-tose	Mal-tose	Man-nite	Raffi-nose	Saccha-rose	Salicin	Starch
1	A C	A C	A C	A C	—	—	A C	A C	—
2	A C	A C	A C	A C	—	—	A C	A C	—
3	A	A C	A	A C	—	—	A C	A C	—
4	A	A C	A	A C	—	—	A C	A C	—
5	A C	A C	A C	A C	—	A	A C	A C	—
6	A C	A C	A C	A C	—	—	A	A C	A C
7	A C	A C	A C	A C	—	—	A C	A C	A C
8	A C	A C	A C	A C	—	—	A C	A C	A C
9	A	A C	A C	A C	—	—	A C	A C	A
10	A C	A C	A C	A C	—	—	A C	A C	A C
11	A C	A C	A C	A C	—	—	A C	A C	A
12	A C	A C	A C	A C	—	—	A C	A C	A
13	A C	A C	A C	A C	—	—	A C	A C	A
14	A C	A C	A C	A C	—	—	A C	A C	A
15	A C	A C	A C	A C	—	—	A C	A C	A
16	A C	A C	A C	A C	—	—	A C	A C	A
17	A C	A C	A C	A C	—	—	A C	A C	A
18	A C	A C	A C	A C	—	—	A C	A C	A
19	A C	A	A C	A C	A	—	A C	A	A
20	A C	A C	A C	A C	A	—	A	A C	—
21	A	A C	A	A C	—	—	A C	—	A

A = acid to litmus.

C = coagulation within 7 days.

In neutral red and methylene blue milk no decolorization was observed. Coagulation was produced by all strains except strains 3, 4, 9 and 21.

According to Holman's¹ classification of hemolytic streptococci, 18 strains correspond to *Streptococcus pyogenes* (ferment lactose and salicin but not mannite); two strains to *Streptococcus infrequens* (ferment lactose, salicin and mannite), and one strain to *Streptococcus anginosus* (ferment lactose but not mannite or salicin). Broadhurst² found no *anginosus* but 4 strains of *Streptococcus infrequens* and 7 strains of *Streptococcus pyogenes* in the throat of 12 dogs.

¹ Jour. Med. Research, 1918, 34, p. 377.

² Jour. Infect. Dis., 1915, 17, p. 277.

My results indicate that *Streptococcus pyogenes* can be roughly divided into two fairly even groups depending on the reaction in the starch medium (see table); whether this is of any significance remains undetermined.

Only three of the pyogenes strains were nonpathogenic for mice, and two of these were starch fermenters. The only raffinose fermenter was pathogenic. Both of the strains of *Streptococcus infrequens* were nonpathogenic. The one *Streptococcus anginosus* was pathogenic.

A serum for the study of the antibodies of the hemolytic streptococci was obtained by the intraperitoneal inoculation of chickens at 5-day intervals with the pyogenes strain. One chicken was immunized over a period of four months increasing the amount injected so that at the last inoculation it received the organisms from 3 liters of ascitic broth. The agglutinins reached their highest titer during the 3rd month of immunization. At that time a dilution of 1:640 agglutinated the homologous strain; afterward the titer dropped so that only a dilution of 1:160 agglutinated. This same serum agglutinated the *infrequens* streptococcus in a dilution of 1:40 at the 4th month, which was the highest titer it ever reached. The dog *anginosus* streptococcus as well as human strains of pyogenes, *infrequens* and *anginosus* never agglutinated in higher dilution than 1:10.

The immune chicken serum fixed complement completely with *anginosus* as antigen, not at all with *infrequens*, and only partially with the homologous pyogenes strain. There was no fixation with the human strains.

The opsonins were increased definitely above normal for the homologous strain (index 4.3), for a human pyogenes strain (index 3.5), but not for *anginosus* and *infrequens* strains.

SUMMARY

Most of the hemolytic streptococci in the throat of the dogs studied were of the pyogenes type. No close relationship could be demonstrated between the groupings of the dog streptococci according to the results of fermentation tests and the results of tests with antistreptococcus chicken serum.